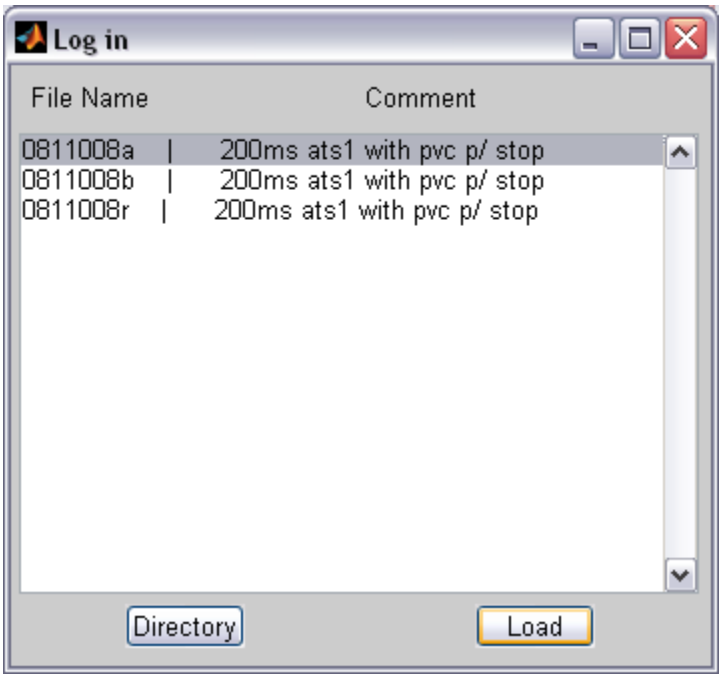
1) Whole heart lab

a. Students will load their data file by typing “zeng” into the matlab command prompt.

b. Using the directory button, locate your file so that the file name and associated comments are displayed in the “Log in” window as shown on the right.



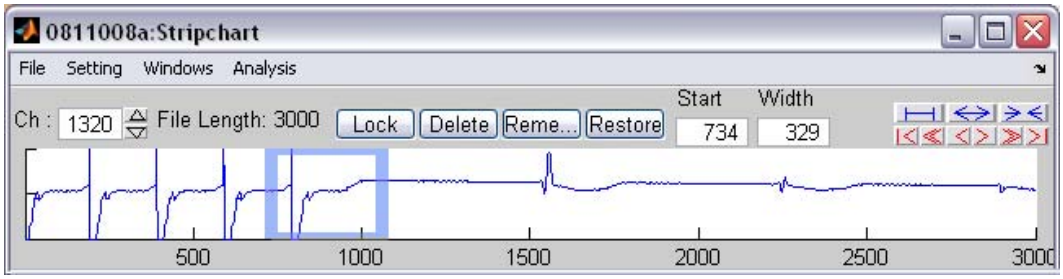
c. Select your file in the “Log in” window and press the “Load” button.

d. The “Stripchart Window” will open. See below.

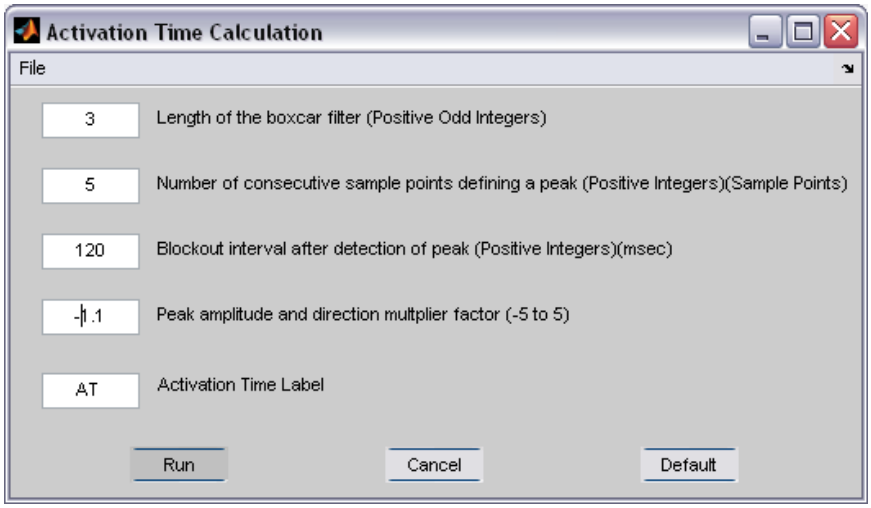
e. Type 1320 into the “channel” edit textbox. This is the ECG channel.

f. Using the mouse, highlight a complete QRS-T complex making sure to start at least 10 msec before the QRS complex.

g. Select “Analysis->Activation Time” from the drop down menu.

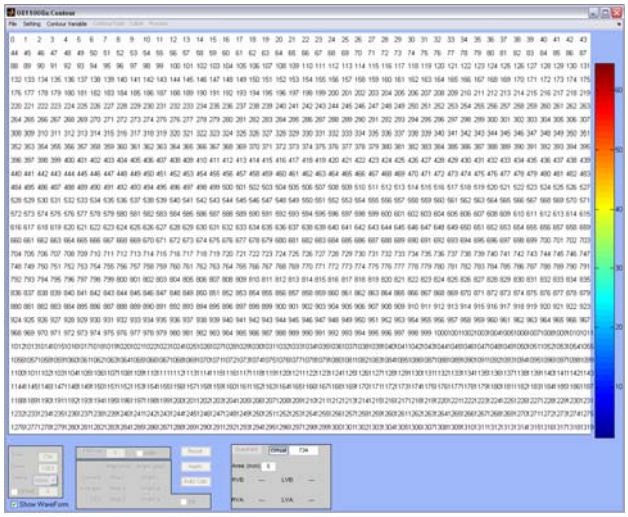


h. Make sure the “Peak amplitude and direction multiplier factor” is set to -1.1 as shown below.



i. Hit run.

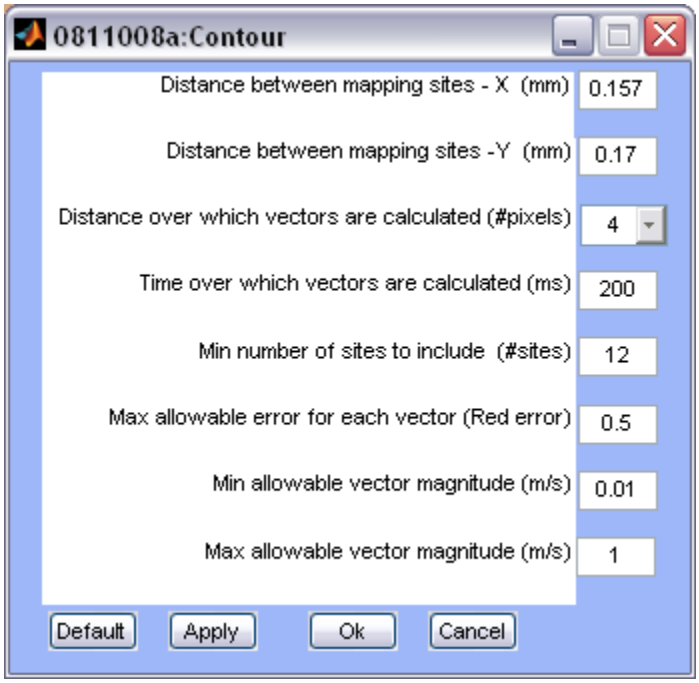
j. Go back to the Stripchart and select “Windows->Contour” from the dropdown menu which will open the “Contour Window” shown below.



k. Select “Contour Variable->AT.” This displays a pixel intensity corresponding to the time of activation within a single pixel.

l. Select “Setting->Vector Parameters.” All parameters should follow the values below. This program tells the conduction velocity algorithm the spacing between pixels and the magnification used to obtain the image. It also tells the program how many pixels to look at before assigning a vector.

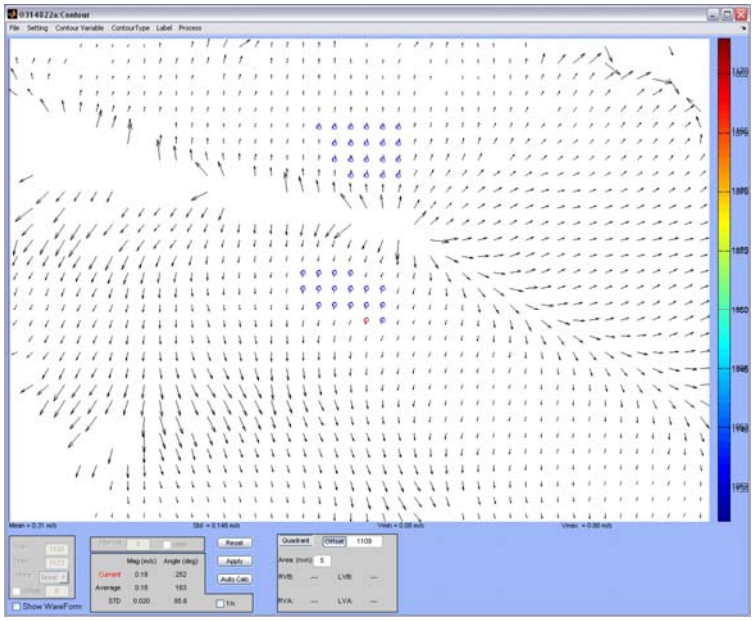
m. Hit Apply.



n. Your “Contour” window will now be filled with many arrows.

o. Select “Label->Channels” to turn the channels off so you can see the vectors clearly.

p. Click on the base of 5 to 15 vectors that are along the short axis of the ellipse as shown below. Note the average and standard deviation of the vector magnitudes. This is your tranverse conduction velocity measurement.

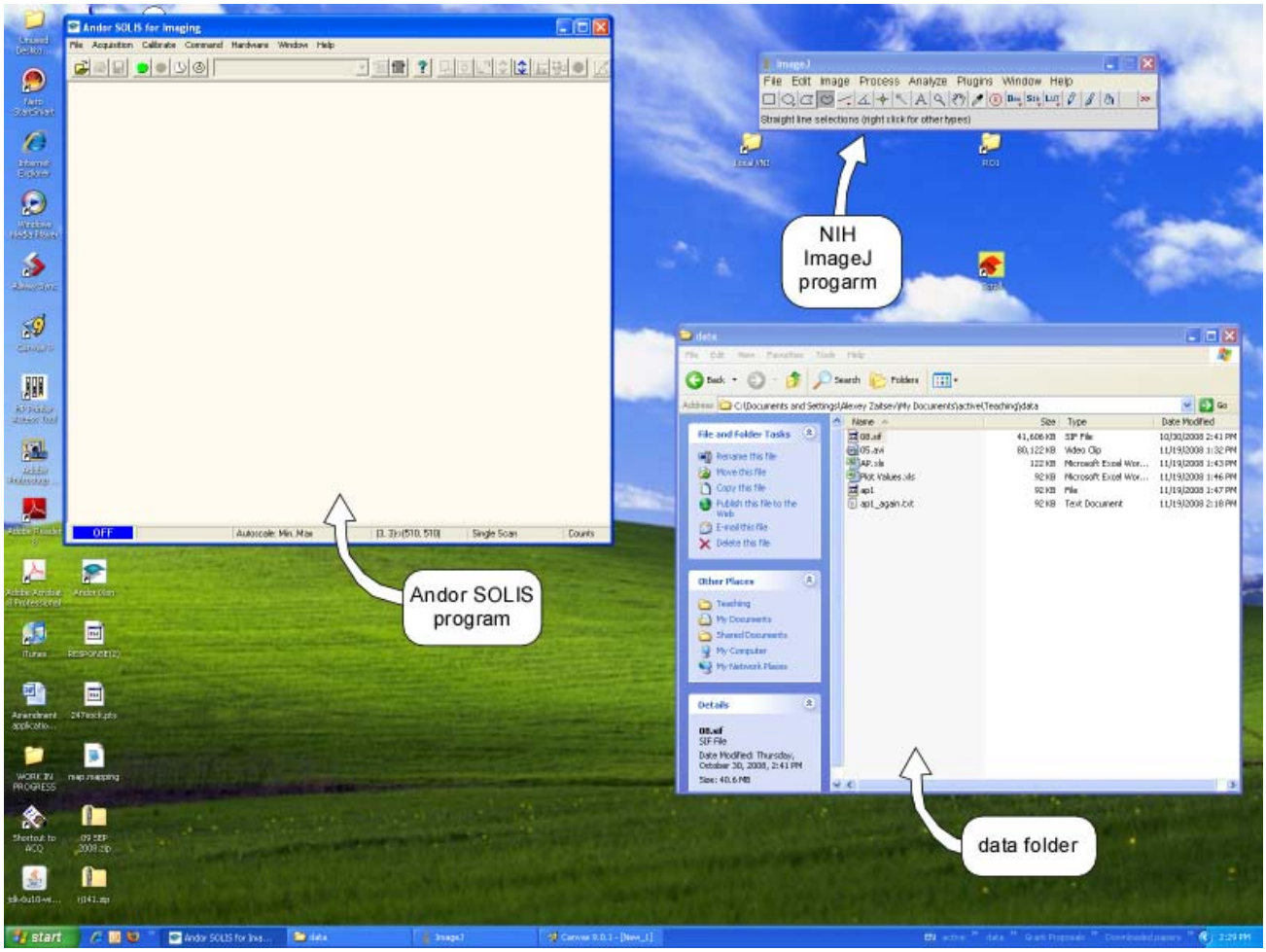


q. Hit Reset.

r. Click on the base of 5 to 10 vectors that are along the long axis of the ellipse. Note the average and standard deviation of the vector magnitudes. This is your longitudinal conduction velocity.

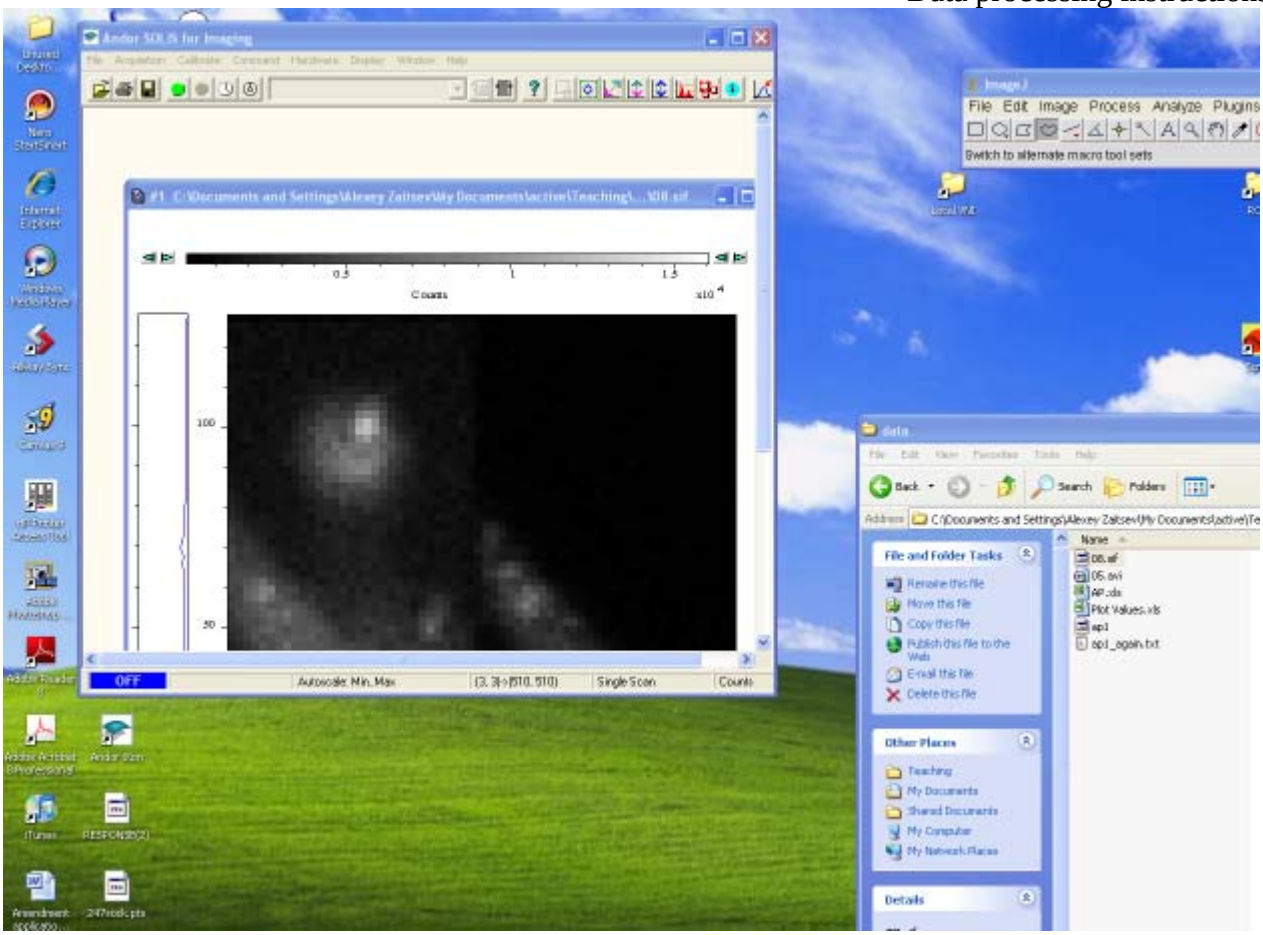
2) Single cell lab

You will need two programs, Andor SOLIS and NIH ImageJ. These programs will be pre-opened by the instructor along with the “data” folder.



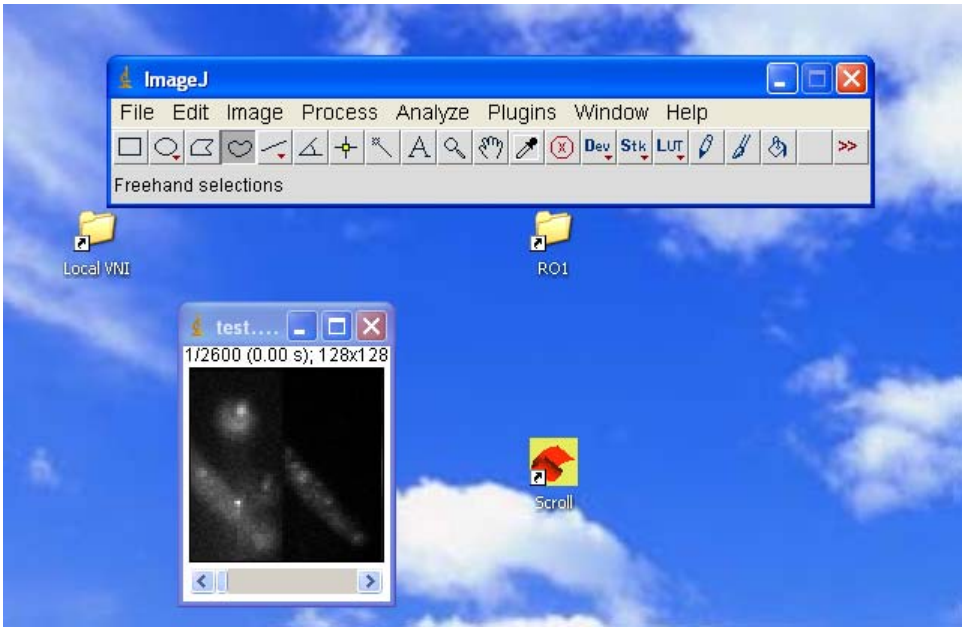
a. Copy your data (SIF files) to the “data” folder.

b. Load a SIF file with the movie of voltage and Ca-sensitive fluorescence recorded from a single cell in the SOLIS program by “drag-and-drop” operation. A window with a movie will appear in SOLIS window:

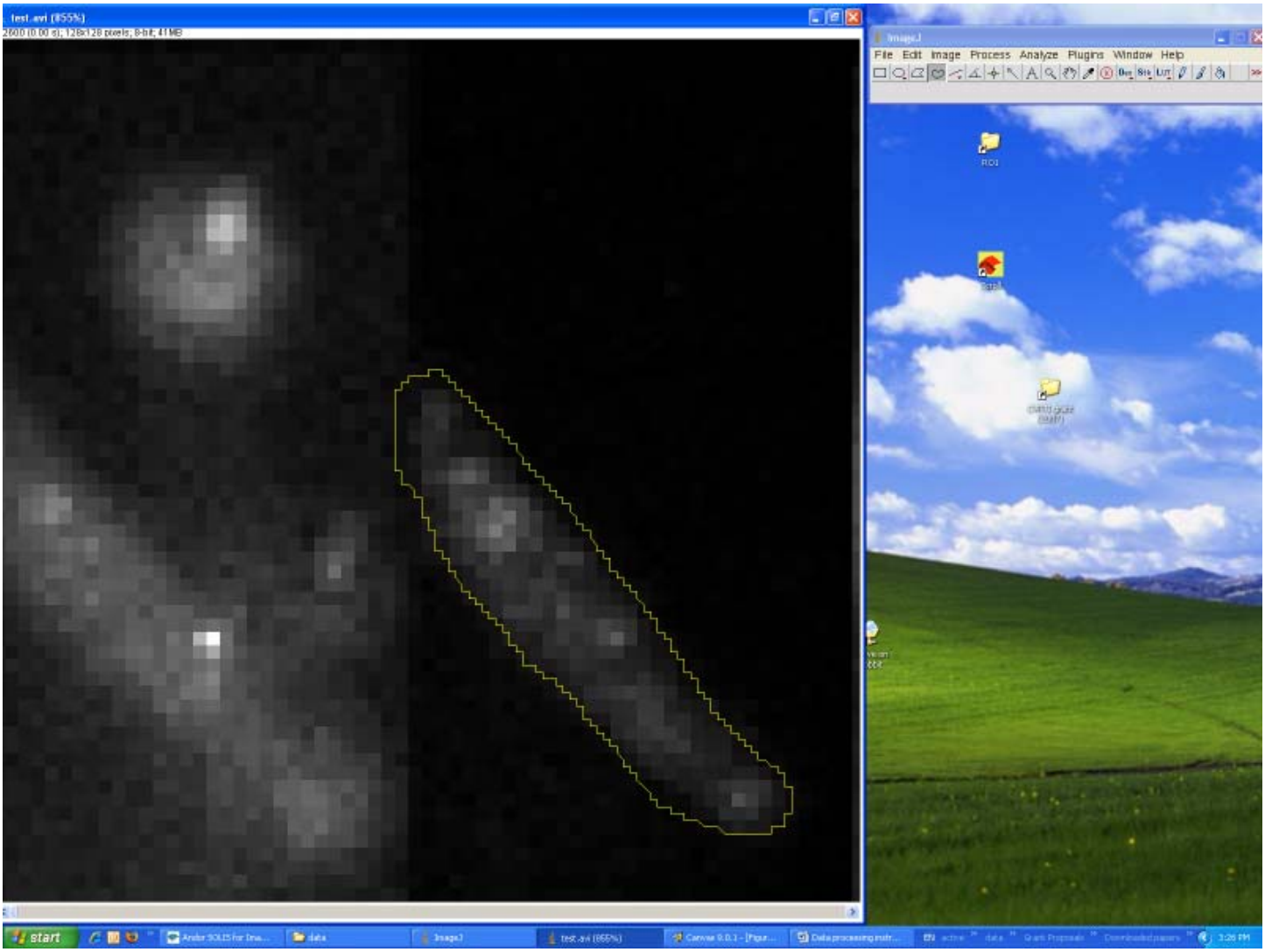


c. In SOLIS, select menu item File/Export As. “Export As” dialog window will appear. Select “avi files” from the drop-down file format menu. Click “save”. The “AVI Export” dialog window will open. Click “OK” to close that window. Wait 2-3 minutes for SOLIS to create the AVI file.

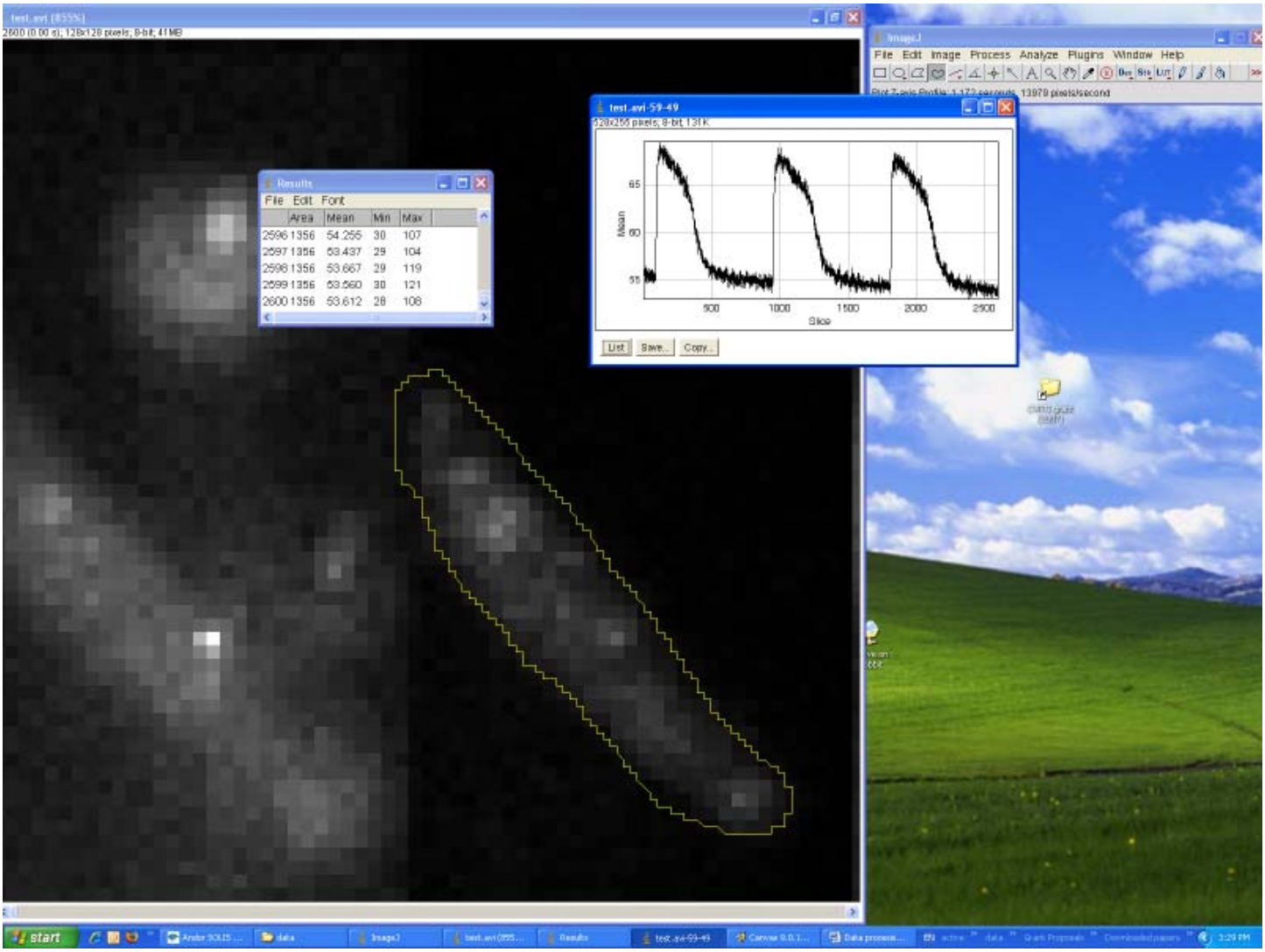
d. To read the AVI file in ImageJ, select menu item File/Open in ImageJ. “Open” dialog window will appear. Select the AVI file of interest. The “AVI Reader” dialog window will open. Check “Convert to Grayscale” checkbox. Click “OK” button. ImageJ will open the AVI file in a separate window:



e. Pick the “Freehand selections” tool from the ImageJ toolbar and select the area containing the cell image. Note that the movie can have more than one cell image. Also, there can be cell debris or other bright objects which are basically garbage. Ask the instructor if you are not sure how to select the cell.



f. To average the signal over the cell area, go to menu item Image/ Stacks/Plot Z-axis profile. ImageJ will create a stack of statistical values calculated for the selected area in all frames of the movie:



g. We are interested only in the average value versus time (time series). In order to save the time series, click “Save” button in the window displaying the time series. A “Save As text” dialog box will appear. Type in the file name, add extension “.txt” and save the text file for subsequent analysis at home. The file contains two columns (frame number and the average level of signal) separated by TAB character. You can use software of your choice (e.g., Matlab, Excel, etc.) to load this text file and analyze it at home.

h. Repeat the steps above for as many original data files and cell images as needed.